ESTROGEN ACTIVITY IN POSTMENOPAUSAL WOMEN*

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AN oversimplified view of the menopause is that at the cessation of ovarian activity and the drop of estrogen production, involution occurs in the genital tract, vasomotor phenomena give rise to hot flush symptoms, and degenerative changes in various organs and systems are accelerated. These changes include osteoporosis, arteriosclerosis, and atrophy of the skin. According to this view the menopause is a deficiency state, similar to diabetes or hypothryoidism. Its treatment, therefore, should be estrogen replacement, which would retard or prevent the aging and degenerative process. The overenthusiastic proponents of "estrogen forever" claim with this treatment not only the relief of flushes and atrophic vaginitis, but also rejuvenation and the cure of headaches, depression, and fatigue.[31] However, the difficulties encountered in the diagnosis and treatment of menopausal symptoms have raised many questions about the exact role of endogenous and exogenous estrogen in the menopause.

HOW IS ESTROGEN EFFECT BEST DETECTED?

Chemical tests for estrogen in urine or plasma are complicated, expensive, and subject to individual variations; therefore maturation of the vaginal epithelium has been the most widely used criterion for estrogen effect both in animal bioassays and in humans. The vaginal smear

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test is simple and can be done repeatedly without discomfort to the patient. The vaginal smear method, however, cannot be used to measure estrogen, since there is no quantitative correlation between the maturation index and the amount of estrogen.[30] In addition, the vaginal smear does not always reflect symptoms.[12]

Do All Postmenopausal Women Have Estrogen Deficiency?

The clinical observation of postmenopausal women indicates that fewer than half complain of symptoms. Vaginal cytology studies made by many investigators show that many postmenopausal women have some estrogen effect. In a survey by Struthers, 58% of the smears taken from women following natural menopause and 51% after oophorectomy showed a moderate to high estrogen effect.[37] DeWaard found "estrogenic" smears, those having more than 20% of karyopyknotic cells, in 9% of postmenopausal women.[5] Other studies by Masukawa[13] and Meisels[15] indicate estrogen activity with moderate effects in approximately 40% of postmenopausal women and high values in 9 per cent. It is not known, however, whether the women with estrogen effect evident on vaginal smears have less vasomotor disturbance, osteoporosis, and arteriosclerosis.

What is the Source of Estrogen in Postmenopausal Women?

The source of postmenopausal estrogen is poorly understood and largely speculative.

Histochemical studies by Novak and his colleagues[20] demonstrated that 50% of postmenopausal ovaries contain enzymes such as 3-β-ol dehydrogenase and DPN diaphorase; these enzymes are necessary for the metabolism of estrogen. Plotz et al.[22] observed the production of androgens and estrogens when microsomal and soluble fractions of postmenopausal ovary were incubated with radioactive steroid precursors. Since there was no correlation between the ovarian findings and estrogenic stimulation of the endometrium, he postulated that the C-19 androgenic compounds found in the ovary may be aromatized to estrogens in the skin or other organs. The finding of estrogen effect after oophorectomy led to the belief that the source might be extraovarian.[27] The adrenal gland, which has all the enzymes necessary for synthesis of steroids, has been frequently cited as the most probable extraovarian site of estrogen production.

What Are the Effects of Estrogen Deficiency in Various Organs?

The response to estrogen deprivation in other parts of the genital tract is less specific than in the vaginal epithelium.

Endometrium in postmenopausal women presents various degrees of involution, total atrophy with cystic dilation of the glands, or a pattern similar to proliferation and hyperplasia, or polyps.

The effect on skin, arteries, and bone is difficult to estimate in quantitative terms. Recently an attempt was made to quantitate the osteoporotic changes by means of x rays[15] and the vibratory density of the ulna.[25]

Is Estrogen Deficiency Harmful?

The common menopausal phenomena, such as atrophy of the gentalia and skin, and osteoporosis, cause discomfort but do not influence life expectancy. On the other hand arteriosclerosis, which has been linked with the menopause, may pose a real threat. Coronary disease is less common in women before menopause than in men of similar age.[6,7,17,23] It increases after the menopause and by the seventh decade becomes equally common in both sexes.[29] In women who had a spontaneous premature menopause ischemic heart disease was observed seven times more frequently than in the general population.[28]

An increased rate of atherosclerosis was also found following oophorectomy. Incidence of ischemic heart disease in women who had a bilateral oophorectomy was four times that found in women who had a hysterectomy without oophorectomy.[24] Elevation of serum cholesterol levels as well as increased incidence of heart disease was reported also after oophorectomy.[11,12]

All these observations led to the conclusion that estrogens may delay the atherosclerotic process, perhaps by maintaining the cholesterol/phospholipid ratio at a low level. On the other hand Novak and Williams, in autopsy studies, found no difference in the incidence of coronary disease between castrated and noncastrated women.[19]

Is Estrogen Excess Harmful to Postmenopausal Women?

Some of the harmful effects of estrogen are known, others are suspected. It is known that estrogen therapy in postmenopausal women may cause uterine bleeding, endometrial hyperplasia, and recurrent

TABLE I. MATURATION VALUE (MODIFIED MEISELS)

	Unit value
Superficial cell	1.0
Large intermediate cell	0.6
Small intermediate cell	0.5
Parabasal cell	0.0

Maturation value
% × unit value = maturation value
0- 49 = low
50- 64 = moderate
65-100 = high

growth of fibroids[19,10] The role of estrogen in the genesis of the tumors is not clear. Evidence of unopposed estrogenic activity in women with fundal cancer was observed both in pre- and postmenopausal groups.[18, 26] A large number of endometrial cancers have been observed in postmenopausal women who were receiving estrogen therapy.[10] Experimental uterine cancer has been induced in animals by means of estrogen.[16] The association of endometrial cancer with polycystic ovaries and the estrogen producing granulosa and theca cell tumors of the ovary[4] also support the view that the endometrial tumors may be estrogen dependent.

From all the foregoing findings it is apparent that deprivation of estrogen is not universal in postmenopausal women and that its effect is difficult to assess. The dangers of hypoestronism are balanced by the risk of estrogen medication and much further investigation is warranted.

THE CURRENT STUDY

The ongoing study at the New York Medical College has five phases. The first four of these have been under way for several years. The fifth phase has been started recently and a preliminary report of findings is anticipated for next year.

The first step in the study is the evaluation of the vaginal smear as a means of determining estrogen effect. Vaginal smears from 469 chronically ill postmenopausal patients at Bird S. Coler Hospital in New York have been studied to date.

The cytohormonal evaluation is made on smears taken from the lat-

TABLE II. ESTROGENLIKE EFFECTS BY YEARS POSTMENOPAUSE

Years postmenopause	*No. patients*	*Maturation value*					
		High (65-100)		*Moderate (50-64)*		*(Low 0-49)*	
		No.	*%*	*No.*	*%*	*No.*	*%*
1- 9	72	7	10	34	47	31	43
10-19	126	9	7	46	37	71	56
20-29	139	9	7	37	27	93	67
30-39	104	6	6	26	25	72	70
40+	28	2	7	6	21	20	72
Totals	469	33		149		287	

eral wall of the vagina. Cells are stained by the standard Papanicolaou method. For purposes of general comparison, three systems of evaluation are used in this study: the Karyopyknotic Index, the Maturation Index, and the Maturation Value. All slides are read independently by two cytologists, one recording the Karyopyknotic Index and the Maturation Index, and the other calculating the Maturation Value. A complete scan of the slide precedes the differential count of at least 200 cells over several fields. The determination of Maturation Value as suggested by Meisels[15] has been modified slightly. The differential count of cell types (superficial, large intermediate, small intermediate, and parabasal) over a number of fields is made under high-power magnification. The per cent of each cell type is multiplied by its specified value (Table I). The sum of these calculations falls between 0 and 100 and is the Maturation Value, interpreted as being low between 0 and 49, moderate between 50 and 64 and high between 65 and 90.

Results of this phase of the study show an increase in the percentage of women with low values (0-49) from 43% during the first 10 menopausal years to 71% in the fifth decade of postmenopausal years. The percentage of women with high Maturation Value (65-100) remained on the same level throughout the postmenopausal years, and dropped only slightly from 10 to 6% (Table II).

The study has confirmed earlier reports indicating that an estrogenlike effect is observed in vaginal smears from a considerable number of postmenopausal women. A gradual decrease of the Maturation Value with increasing age and postmenopausal years is in general agreement with the findings of others. However, we find a slightly larger shift toward the lower levels and no marked drop after the tenth postmeno-

TABLE III. MATURATION VALUE IN VARIOUS DISEASES

Disease	No. patients	Maturation value: High (65-100) No.	%	Moderate (50-64) No.	%	Low (0-49) No.	%
Heart and vascular	206	22	11	62	30	122	59
Diabetes	78	2	3	29	37	47	60
Central nervous system	61	8	13	23	38	30	49
Arthritis	37	4	11	13	35	20	54
Fractures	20	1	5	4	20	15	75
Renal	9	0	0	4	44	5	56
Liver (cirrhosis)	14	1	7	9	64	4	29
Others	44	2	5	12	27	30	68
Total	469						

pausal year as reported in other series. It must be reiterated, however, that our patients are chronically ill and that those in the other reported series were presumably in good health or their health status was unspecified.

The second phase of the study is the correlation of the observed estrogenic values with various diseases and medications. Maturation Value by disease groups were found to be mostly within the same percentage range (Table III).

Because of fairly uniform results, the frequent overlapping of diseases, and the small numbers of patients in each category, no statistical analysis was made. The only groups which deserve some comment are those that had diabetes or cirrhosis of the liver, and patients on digitalis therapy.

In 78 diabetics, the percentage of high Maturation Value was less than that in the general population, while the percentage in the moderate and low classifications were within the same statistical range.

Diabetes has been associated with high estrogen effects, hypertension, and obesity. It has also been reported that the incidence of endometrial carcinoma, which is believed to be dependent on estrogen is higher in this group. We do not find elevated Maturation Value in diabetics.

In 11 cases of cirrhosis of the liver the finding of small numbers of women with low Maturation Value is not surprising, since it is generally believed that the conjugation of estrogen is impaired by this disease.

TABLE IV. MATURATION VALUES IN PATIENTS ON DIGITALIS THERAPY AND ALL OTHERS

	Maturation value					
	High (65-100)		*Moderate (50-64)*		*Low (0-49)*	
	No.	*%*	*No.*	*%*	*No.*	*%*
Digitalis Rx*	9	8	34	31	68	61
Others	25	7	114	32	219	61

*On therapy for at least one year.

Among the 111 patients on digitalis therapy (Table IV) the incidence of high Maturation Value is not significantly greater than the average. All patients within this group had been receiving digitalis for more than one year and several for more than five. This finding refutes our previous report, which was based only on 32 cases.

The reports in the literature of the effects of digitalis on the vaginal mucosa are controversial. Some investigators have reported a significant increase in maturation of the vaginal epithelium of women who were taking digitalis.[20] This reaction is explained by the similarity in molecular structure of the active principle of digitalis glycoside (aglycone) and that of estrogen. Conversely, Gordon and his colleagues[8] found no increase in maturation in a controlled study of nine postmenopausal women on digitalis therapy. However, his patients were on therapy for "at least one month" and, in some cases, for only one month. While estrogen therapy is reflected by an almost immediate change in the vaginal mucosa, our findings suggest that digitalis requires a much longer period of time.

The third step is a correlation of the cytohormonal findings with the changes in the uterus. For this purpose the uteri obtained at autopsy from previously screened patients are examined. To date the number of prospectively studied women is small, but certain trends have been observed.

In patients with atrophic smears, marked atrophy of the endometrium was seen. In women with moderate estrogen effect the endometrium showed some evidence of proliferative activity.

The fourth step in our investigation is an attempt to find a practical means of evaluation of the endometrial status without resorting to curettage or biopsy. Cytological material from the uterine cavity is

TABLE V. CYTOCHEMISTRY OF ENDOMETRIAL WASHINGS

	ALP	*ACP*	*Fluorescence*
Atrophy	O	+*	+
Proliferative	++	O	++
Hyperplastic	++	O	++
Carcinoma	O, or ++	O*	+++

*Positive in macrophages.

examined for this purpose. The findings are then compared with histological characteristics on tissue material. The cell samples are obtained by irrigating the uterine cavity with normal saline through a polyethylene catheter. Since the morphologic characteristics of the endometrial cells are not sufficiently defined, cytochemical staining was used. Endometrial cells are stained for alkaline and acid phosphatase by the method of Burstone and Barka[2, 3] and with acridine orange for fluorescence study. Tissues from hysterectomy, or curettage specimens, are stained in the same manner. The intensity of alkaline and acid phosphatase reaction and fluorescence are then correlated with morphology both in irrigation and tissue specimens. Patients in the proliferative phase with postmenopausal atrophy, cystic hyperplasia, and adenocarcinoma have been evaluated.

The results of cytochemical findings are summarized in Table V. The intensity of alkaline phosphatase was found to rise in proportion to the proliferative activity. Acid phosphatase was more intense in involuting than in growing tissue. In cases of carcinoma the fluorescence was strong in the histiocytes, but negative in epithelial cells. Good correlation was observed between the findings on irrigation and tissue samples.

It is hoped that the cytochemical method when perfected will become useful in evaluating the activity of endometrial epithelium in women receiving estrogen supplement and will serve as a guide to treatment.

The fifth step in the study of the estrogen effect in postmenopausal women is in the investigation of the role of endogenous estrogen in the prevention of atherosclerosis. For this purpose estrogen effect is esti-

mated from vaginal smears of patients confined to a chronic disease hospital. In the event of a death, the degree of atherosclerotic change is evaluated and is correlated with the estrogen index. A significantly low incidence of atherosclerosis found in women with signs of high estrogen activity will indicate that endogenous estrogen has indeeed a protective action.

The study will attempt to present additional evidence in favor of or against the concept that estrogen protects blood vessels. It is hoped that the results will allow more selective and judicious use of exogenous estrogen.

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